

Claims

1. An isolated nucleotide sequence responsible for the tomato *high pigment 1* (*hp-1*) phenotype, wherein said sequence comprises an altered tomato *DDB1* gene sequence or fragment thereof, wherein said the alteration in said altered sequence or fragment comprises an A-to-T transversion at nucleotide 931 of said *DDB1* gene sequence.
2. The isolated nucleotide sequence according to claim 1, wherein said sequence comprises the sequence defined as SEQ ID NO:1 in the sequence listing.
3. The isolated nucleotide sequence according to claim 1, wherein said sequence comprises a fragment of SEQ ID NO:1, and wherein said fragment comprises nucleotide 931 of the *DDB1* gene sequence.
4. An isolated nucleotide sequence responsible for the tomato *high pigment 1<sup>w</sup>* (*hp-1<sup>w</sup>*) phenotype, wherein said sequence comprises an altered tomato *DDB1* gene sequence or fragment thereof, wherein said the alteration in said altered sequence or fragment comprises a G-to-A transition at nucleotide 2392 of said *DDB1* gene sequence.
5. The isolated nucleotide sequence according to claim 4, wherein said sequence comprises the sequence defined as SEQ ID NO:2 in the sequence listing.
6. The isolated nucleotide sequence according to claim 4, wherein said sequence comprises a fragment of SEQ ID NO:2, and wherein said fragment comprises nucleotide 2392 of the *DDB1* gene sequence.

7. A method for detecting the presence of the *hp-1* mutation in a plant, comprising the steps of isolating the genomic DNA from said plant, amplifying a gene fragment containing said *hp-1* mutation from said genomic DNA by use of a PCR technique and determining the presence of said *hp-1* mutation in said genomic DNA.

8. The method according to claim 7, wherein the presence of the *hp-1* mutation is determined by the use of a pyrosequencing technique, and wherein the sequence data obtained from said technique is compared with the sequence defined in SEQ ID NO:1.

9. The method according to claim 7, wherein the plant in which the presence of the *hp-1* mutant is being detected is of the species *Lycopersicon esculentum*.

10. A method for detecting the presence of the *hp-1<sup>w</sup>* mutation in a plant, comprising the steps of isolating the genomic DNA from said plant, amplifying a gene fragment containing said *hp-1<sup>w</sup>* mutation from said genomic DNA by use of a PCR technique and determining the presence of said *hp-1<sup>w</sup>* mutation in said genomic DNA.

11. The method according to claim 10, wherein the presence of the *hp-1<sup>w</sup>* mutation is determined by the use of a pyrosequencing technique, and wherein the sequence data obtained from said technique is compared with the sequence defined in SEQ ID NO:2.

12. The method according to claim 10, wherein the plant in which the presence of the *hp-1<sup>w</sup>* mutant is being detected is of the species *Lycopersicon esculentum*.

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13. Use of the method according to either claim 7 or claim 10 as a means of post-control in seed production.

14. A method for the determination of the presence of two different photomorphogenic mutations in a plant, wherein one of said mutations is either the *hp-1* or the *hp-1<sup>w</sup>* mutation, comprising detecting the presence of a photomorphogenic mutation other than the *hp-1* or the *hp-1<sup>w</sup>* mutation by either genotypic or phenotypic selection means, and detecting the presence of the *hp-1* or the *hp-1<sup>w</sup>* mutation by means of the method according to either claim 7 or claim 10.

15. The method according to claim 14, wherein the phenotypic selection means for determining the presence of the non-*hp-1*, non-*hp-1<sup>w</sup>* photomorphogenic mutation comprises germinating seeds obtained from the plant in which the presence of the mutations is being determined in a temperature controlled chamber, under a yellow plastic screen that is opaque to light having a wavelength less than 500nm, and selecting non-etiolated seedlings.

16. A method for preparing double-mutant lines of *Lycopersicon esculentum* having genotype *hp-1/hp-1 p/p*, wherein *p* represents any recessive photomorphogenic lycopene-enhancing mutation that is genetically unlinked to the *hp-1* mutation, said method comprising the steps of:

- a) cross-hybridization of a homozygous *hp-1/hp-1* line or plant with a homozygous *p/p* line or plant to yield double heterozygous *hp-1/+ p/+ F<sub>1</sub>* plants;
- b) self-crossing of the *F<sub>1</sub>* plants obtained in step (a) in order to yield *F<sub>2</sub>* seeds;

- c) identification of double homozygous plants  $hp-1/hp-1\ p/p$  by means of the application of the method defined in claim 7 and a method for detecting the presence of the  $p$  mutation;
- d) self-crossing of the double homozygous plants identified in step (c) to generate  $F_3$  seeds, and germination of said seeds.

17. The method according to claim 16, wherein mutation  $p$  is the  $dg$  mutation.

18. The method according to claim 17, wherein the determination of the presence of the  $dg$  mutation in step (c) of the method is performed using the marker for the  $dg$  mutation disclosed in co-owned, co-pending application PCT/IL03/00023.

19. A method for preparing double-mutant lines of *Lycopersicon esculentum* having genotype  $hp-1^w/hp-1^w\ p/p$ , wherein  $p$  represents any recessive photomorphogenic lycopene-enhancing mutation that is genetically unlinked to the  $hp-1^w$  mutation, said method comprising the steps of:

- a) cross-hybridization of a homozygous  $hp-1^w/hp-1^w$  line or plant with a homozygous  $p/p$  line or plant to yield double heterozygous  $hp-1^w/+ p/+ F_1$  plants;
- b) self-crossing of the  $F_1$  plants obtained in step (a) in order to yield  $F_2$  seeds;
- c) identification of double homozygous plants  $hp-1^w/hp-1^w\ p/p$  by means of the application of the method defined in claim 10 and a method for detecting the presence of the  $p$  mutation;
- d) self-crossing of the double homozygous plants identified in step (c) to generate  $F_3$  seeds, and germination of said seeds.

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20. The method according to claim 19, wherein mutation *p* is the *dg* mutation.

21. The method according to claim 20, wherein the determination of the presence of the *dg* mutation in step (c) of the method is performed using the marker for the *dg* mutation disclosed in co-owned, co-pending application PCT/IL03/00023.